EXCRETION OF MINERALS AND NITROGEN METABOLITES FOLLOWING EXPOSURE TO INCREASED AIR PRESSURES (2 or 7 ATA)

by

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SUMMARY PAGE

THE PROBLEM

To further evaluate the long-term effect of pressure and decompression on mineral and nitrogen metabolism, distribution and loss in men subjected to standard Navy diving procedures.

FINDINGS

A reduction in urine output, with a concomitant decrease in mineral and nitrogen metabolite excretion, has been observed during the first 24 hours following exposure to 2 and 7 ATA of compressed air. During the succeeding three days, volume, mineral, and electrolyte components progressively returned to control values while uric acid, creatinine and ketosteroids continued to remain depressed.

Increasing excretion of hydroxyproline following exposure to 7 ATA suggests latent responses of cartilage and bone to this stress.

APPLICATION

These and previous studies in rats indicate that diving, with or without clinical manifestation of bends, apparently results in biochemical consequences of several days duration. Therefore, careful consideration should be given to providing sufficient recovery time between pressure exposures in order to minimize possible long term detrimental effects on the health of divers.

ADMINISTRATIVE INFORMATION

This investigation was conducted as part of Bureau of Medicine and Surgery Research Work Unit MF51.524.014-9016BA9K. The present report is number two on this work unit. It was submitted for review on 20 September 1973, approved for publication on 10 October 1973 and designated as NavSubMedRschLab Report No. 765.

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ABSTRACT

Urinary minerals, electrolytes, nitrogen metabolites, and steroids were measured in Navy divers following exposure to air at 2 and 7 ATA for 45 minutes.

Total urinary excretion of osmoles, sodium, potassium, calcium, phosphorus, urea nitrogen, uric acid, creatinine, keto- and ketogenic steroids were depressed below control values during the first day following exposure to either 2 or 7 ATA. The mineral and electrolyte changes are related to reduction in urine volume, which may result from an anti-diuretic hormone (ADH) mediated response to a hypovolemia induced by dysbaric stress. Although a general rebound effect began to occur by the second post-dive day, a continued depression of uric acid, creatinine, and ketosteroids throughout four days of observation suggests a prolonged period of recovery from diving.

Increasing excretion of hydroxyproline for several days by those men exposed to 7 ATA, implies a latent response in the metabolism of cartilage and bone to this stress.

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INTRODUCTION

Alterations in blood and urinary electrolytes during periods spent under increased pressure and during decompression from such pressures have been frequently reported.[1,3,4,12,14,15] These changes have been attributed to hyperventilation and fluid shifts, alterations in membrane permeability related to inert gas narcosis, or increased corticosterone and/or aldosterone production. However, observations of changes of minerals and electrolytes in man during a period of several days post-dive have not been reported to our knowledge.

Studies in rats for up to five days following severe decompression stress have shown that mineral and electrolyte changes occur in serum within one hour and in urine within one day post-dive and are followed by a second period of changes after three days.[6] These responses have been attributed to adrenal cortical response and post-decompression hemoconcentration.

Since severe decompression injury appears to result in prolonged mineral and electrolyte responses, consideration should be given to the effect of "safe", apparently non-injurious, decompression on possible latent changes in mineral distribution and/or loss. The purpose of this study is to investigate mineral changes that may occur in Navy divers following safe decompression from 2 and 7 ATA air-breathing simulated dives.

MATERIALS AND METHODS

Ten qualified Navy divers participated in these studies. The simulated dives commenced at 0830 daily, Tuesday through Thursday. The divers for each depth were selected at random and on only four occasions did a new dive occur for a particular diver within 4 days of his preceding dive. The overlap values were eliminated from the study. The design of the experiment randomized the order in which the 2 and 7 atmosphere exposures were performed.

Compression was made at the rate of 75 ft./min. Time on the bottom at each pressure was 40-45 minutes, and decompression was performed according to the Navy Standard Diving Table for Exceptional Exposure (210 ft. for 50 min.).[17] Since it was desired that the men not be aware whether they had been subjected to 2 or 7 ATA, those who had been exposed to 2 ATA were decompressed for a length of time equal to that used for those exposed to 7 ATA.

Since the protocol for the dives involved visual and mechanical performance tests which might have been impaired by biological sampling procedures, no blood or urine samples were obtained during the dives. After emptying the bladder prior to the beginning of the dive, each man collected 24 hour urine samples into concentrated HCl for four consecutive post-dive days. Thus, the urine sample for the first day post-dive sample also contained the

urine produced during the period of the dive.

Prior to these studies each man collected two 24 hour urine specimens to serve as pre-dive controls. During the studies the men refrained from eating ice-cream, gelatin desserts, or soft candy inasmuch as these materials contain large amounts of hydroxyproline.

Total calcium and inorganic phosphorus were determined in the Technicon Autoanalyzer using the simultaneous micromethod N-82 I/II. Sodium and potassium were measured in an Instrumentation Laboratory Model 343 flame photometer. Urine osmolarity was determined in a Fiske Osmometer Model G62. The method of Hosley et al[7] was used to determine the hydroxyproline levels. Urea nitrogen, creatine, and uric acid measurements were made by Autoanalyzer techniques N-13b and N-38a.

Following preliminary procedures consisting of hydrolysis, two washings with 20% NaOH and two washings with ether saturated water, ether extracts of urine samples were analyzed for 17-ketosteroids by the automated procedure of Zak et al.[18] The same color development technique was used for ketogenic steroid analyses following preliminary oxidation, hydrolysis and extractions.[16]

RESULTS AND DISCUSSION

The results from analyses of the urine collections, in terms of total excretion during 24-hour periods, are presented as follows: Figure 1 - volume and osmoles; Figure 2 - sodium

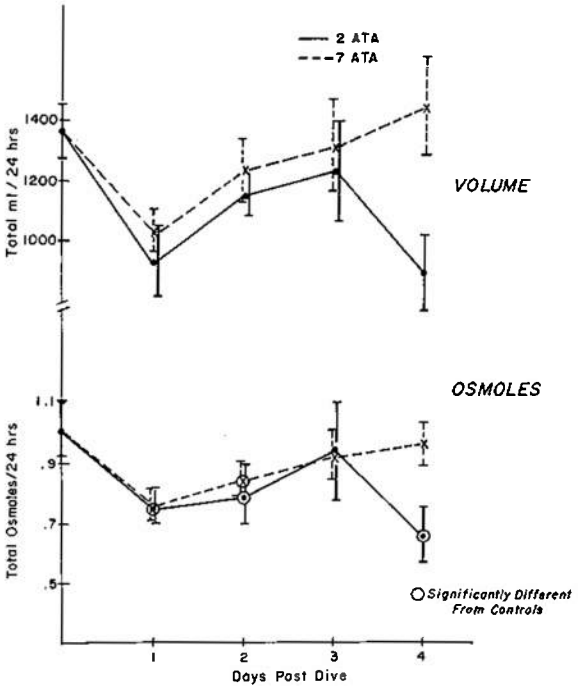

Fig. 1. Total urinary excretion/24 hrs. Volume and osmoles following exposure to air at 2 or 7 ATA. Mean ±SEM.

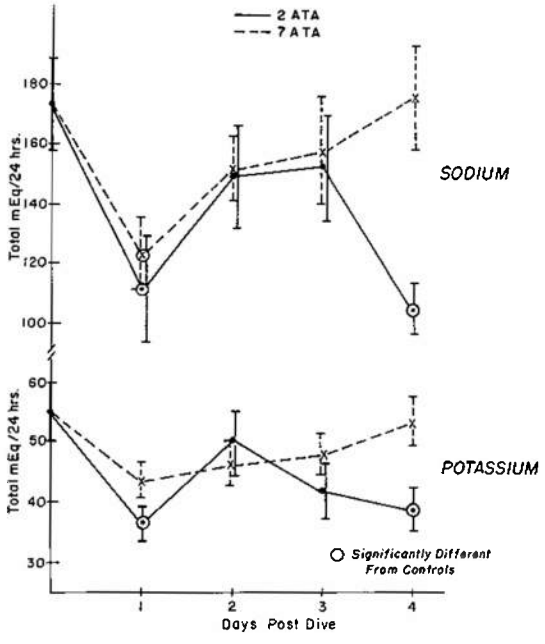

Fig. 2. Total urinary excretion/24 hrs. Sodium and potassium following exposure to air at 2 or 7 ATA. Mean ±SEM.

and potassium; Figure 3 - hydroxyproline, calcium, and inorganic phosphorus; Figure 4 - urea nitrogen, uric acid, and creatinine; Figure 5 - ketosteroids and ketogenic steroids. In Appendix I and II, the data are presented in terms of concentration for each of the above mentioned parameters. Concentration data are presented since these have been demonstrated to exhibit better correlations among the stress-related components of urine then do total excretion data.[16]

During the first day following exposure to air at 2 and 7 ATA, there occurred highly significant decreases in total 24-hour excretion of osmoles, sodium, potassium, phosphorus, urea nitrogen, uric acid, creatinine, keto- and ketogenic steroids. Although the

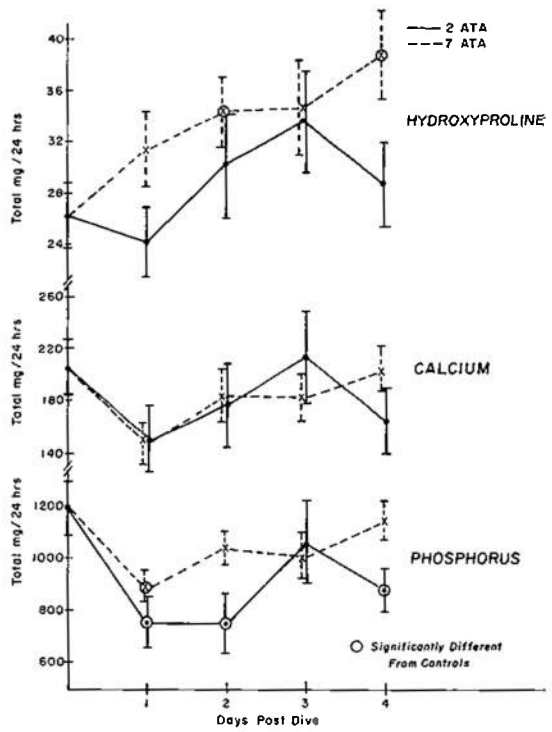

Fig. 3. Total urinary excretion/24 hrs. Hydroxyproline, calcium, and inorganic phosphorus following exposure to air at 2 or 7 ATA. Mean ±SEM.

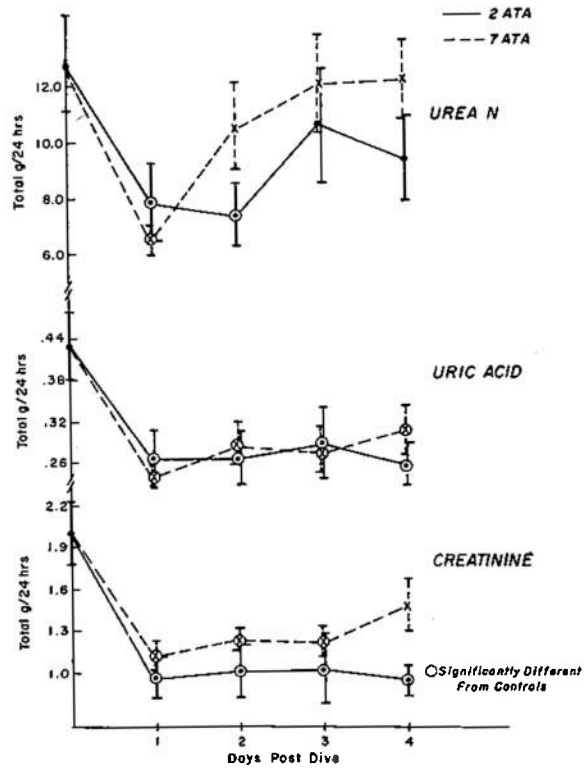

Fig. 4. Total urinary excretion/24 hrs. Urea nitrogen, uric acid and creatinine following exposure to air at 2 or 7 ATA. Mean ±SEM.

decreases were not significant, the output of hydroxyproline as well as calcium and urine volume from those men exposed to two ATA also tended to fall during this time. With the exception of ketosteroid, uric acid, and creatinine excretion, which remained significantly below control levels throughout the study, the other parameters began to return toward control values by the second day post-dive. It is most interesting to note that these responses mirror very closely those observed in rats during several post-dive days following exposure to severe decompression stress.[6]

The generally direct relationship between urine volume and total

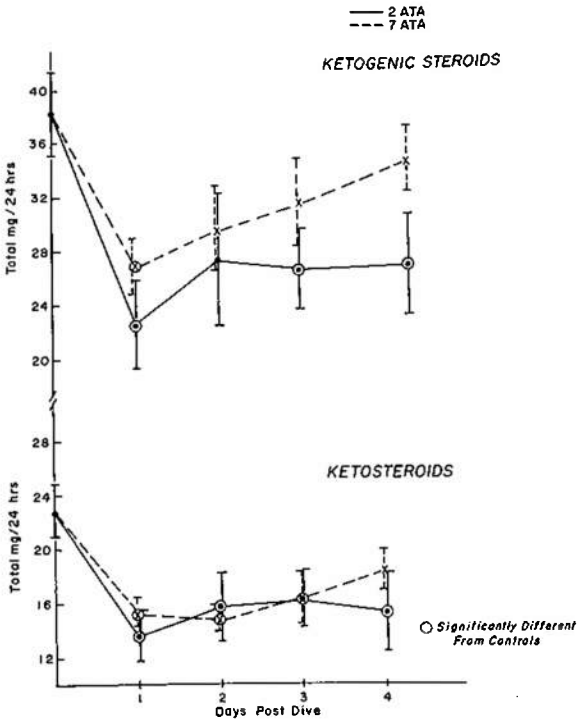

Fig. 5. Total urinary excretion/24 hrs. Ketogenic steroids and ketosteroids following exposure to air at 2 or 7 ATA. Mean ± SEM.

minerals and electrolytes excreted can be noted throughout these studies. This effect is particularly evident on the 4th day after exposure to 2 ATA. The pronounced decrease in volume of urine is sharply reflected in the total excretion of osmoles, sodium, calcium, phosphorus, and urea nitrogen, and, to a lesser extent, of potassium and uric acid. The excretion of these materials when considered on a concentration basis is not significantly different from controls (Appendix I).

It will be observed that the urinary responses to 2 ATA appear to be somewhat greater, and in the cases of phosphorus, uric acid, and ketogenic steroids, of longer duration than the responses to 7 ATA. Again, these responses can be related to variations in the volume of urine excreted and cannot be explained on the basis of individual subject differences since the same divers participated in both exposures. The standard errors of the mean do not support the idea that the fewer number of dives made at 2 ATA than at 7 ATA could account for the observed greater responses and longer recovery periods. It is apparent that in these men, a metabolic response of three or more days duration was triggered by exposure to pressure as little as 2 ATA for 45 minutes.

Excretion of hydroxyproline following exposure to 7 ATA was the only parameter studied which did not decrease on the first post-dive day. Urinary hydroxyproline was somewhat elevated after one day, became statistically greater than controls on the second post-dive day and by the fourth day had risen to an even higher level. The rise in hydroxyproline excretion by the third day following the 2 ATA dives suggests that exposures to even minimal pressures can cause pronounced changes in the metabolism of cartilage and bone.

Calculation of the relationship between sodium and potassium excretion reveals that the only significant change in the Na/K ratio is a decrease that occurs on the first post-dive day following the 7 ATA dives. Since a relative increase in potassium is considered to be an indicator of non-specific stress, this response supports the subjective impressions of the divers that 7 ATA is more stressful than 2 ATA.

Decreases in urine volume following exposure to increased pressure have been previously observed following both short-term dives in rats and saturation dives in man.[1,6] In addition, Arturson and Grotte[2] have reported increased hematocrit and tissue edema in severely decompressed dogs while Jacey et al[8] have concluded that the hemoconcentration they observed in rats exposed to severe decompression stress was related to a plasma deficit. Furthermore, hypovolemia in man during prolonged dives has been reported by Johnson et al.[9] Thus, post-dive decrease in urine volume might be interpreted as a rebound effect from the diuresis observed during periods of increased pressure and/or a compensatory correction of hypovolemia.[1,10,15]

Since urines were collected as 24-hour specimens beginning on the morning of the dives, pressure-induced diuresis could not be observed. It is apparent, however, that the overall effect following exposure to pressure may be a temporary retention of body fluid, an indicator of an augmented ADH supply. Since an increased ADH production results from reduced plasma volume,[13] then one is led to the interpretation that the decrease in urine volume following these dives may have resulted from an ADH mediated response to hypovolemia.

Analyses of nitrogen metabolites indicate minimal tissue damage resulting from exposure of the subjects to the pressures of these experiments. The brevity of the exposures may account for the apparent discrepancy between these results and those of Alexander et al.[1] in which urinary uric acid values increased throughout a 14-day exposure to a normoxic-nitrogen mixture at 4 ATA. Serum enzyme analyses also furnished evidence of tissue damage under these latter circumstances. Although no blood samples which could provide similar enzymic evidence were collected in the present studies, the excretion of uric acid, which in fact was less strongly influenced by urinary volume than any of the other components measured, indicates the possibility of a period of cellular synthesis. This possible synthesis appears to extend for several days after each of the dive schedules. While this speculation is highly tentative, the reduced creatinine excretion might also indicate a similar conclusion. It must be remembered, however, that no tissue damage which would necessitate a period of recovery has been demonstrated.

If it is assumed that the reduced 17-ketosteroid excretion observed during most of the period of these studies is analogous to the decreases reported during periods of stress,[5,11] then the idea is supported that a stress-like syndrome of several days duration follows exposure to increased air pressures.

Certainly the conclusion holds here, perhaps even more firmly than for reduced altitude stress,[5] or for avoidance stress,[11] that "full recovery from multifactor stress apparently requires more time than is needed to induce the stress itself."

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APPENDIX I

EFFECT OF EXPOSURE OF MAN TO AIR AT 2 ATA ON POST-DIVE URINARY EXCRETION. TWENTY-FOUR HOUR COLLECTIONS. MEAN ±STANDARD ERROR OF THE MEAN.

POST DIVE	Vol	Na	K	Hydroxy-proline	Ca	P	Osmolarity	Urea N	Creatinine	Uric Acid	KS*	KGS**
	ml	mEq/l		µg/ml	mg/100ml		mOs/l	g/l			mg/l	
Control												
$\bar{X}$	1367	145.5	46.5	22.4	18.28	103.5	916.2	11.15	1.82	.387	20.60	33.62
SEM	195	10.2	4.4	2.1	2.11	8.8	59.9	.88	.20	.023	2.32	3.41
N	22	22	22	22	22	22	22	13	13	13	22	22
1 Day												
$\bar{X}$	930	144.0	48.5	28.5	17.43	92.6	904.4	8.40	1.20	.308	16.83	28.38
SEM	123	14.5	4.4	2.7	2.73	10.8	60.2	.65	.12	.028	2.26	4.44
N	16	16	16	16	16	16	16	16	16	16	16	16
p								<.05	<.01	<.05		
2 Days												
$\bar{X}$	1167	153.4	47.5	28.6	16.67	76.1	754.9	7.04	1.11	.254	15.09	25.42
SEM	186	14.8	4.5	2.7	1.92	9.2	80.6	1.68	.13	.030	2.11	3.29
N	16	16	16	16	16	16	16	16	16	16	16	16
p						<.05		<.01	<.01	<.01		
3 Days												
$\bar{X}$	1243	134.5	37.5	27.0	17.76	86.0	923.3	8.71	.95	.243	14.69	24.24
SEM	170	16.6	3.9	3.4	2.35	10.2	93.0	.96	.10	.028	2.56	3.01
N	14	13	13	14	13	13	14	13	13	13	14	14
p									<.001	<.001		
4 Days												
$\bar{X}$	908	141.2	46.9	31.7	19.55	109.0	884.4	10.13	1.15	.306	17.74	31.85
SEM	121	16.7	4.1	2.7	2.49	15.4	79.8	1.05	.09	.023	2.87	3.78
N	13	12	12	13	12	12	13	14	14	13	13	12
p				<.02					<.01	<.05		

* Ketosteroids

** Ketogenic steroids

APPENDIX II

EFFECT OF EXPOSURE OF MAN TO AIR AT 7 ATA ON POST-DIVE URINARY EXCRETION. TWENTY-FOUR HOUR COLLECTIONS. MEAN ±STANDARD ERROR OF THE MEAN.

POST DIVE	Vol	Na	K	Hydroxy-proline	Ca	P	Osmolarity	Urea N	Creatinine	Uric Acid	KS*	KGS**
	ml	mEq/l		µg/ml	mg/100ml		mOs/l	g/l			mg/l	
Control												
$\bar{X}$	1367	145.5	46.5	22.4	18.28	103.5	916.2	11.15	1.82	.387	20.60	33.62
SEM	195	10.2	4.4	2.1	2.11	8.8	59.9	.88	.20	.023	2.32	3.41
N	22	22	22	22	22	22	22	13	13	13	22	22
1 Day												
$\bar{X}$	930	125.8	45.9	30.6	14.82	97.1	776.2	6.64	1.21	.257	16.06	28.01
SEM	123	10.7	3.7	1.7	1.33	8.9	65.1	.65	.13	.024	1.53	2.72
N	16	22	22	23	21	23	23	23	22	22	20	23
p				<.01				<.001	<.01	<.01		
2 Days												
$\bar{X}$	1167	135.7	42.1	31.3	15.76	97.7	730.5	7.91	1.11	.238	13.33	25.58
SEM	186	11.7	3.2	2.5	1.31	8.7	65.1	.78	.10	.017	1.30	2.97
N	16	20	20	21	20	21	21	21	21	21	21	22
p				<.02			<.05	<.05	<.001	<.001	<.001	
3 Days												
$\bar{X}$	1243	137.1	40.9	26.0	16.07	88.1	754.7	8.74	1.04	.218	13.44	27.20
SEM	170	13.8	4.0	2.1	1.73	8.1	56.4	.70	.09	.020	1.47	3.51
N	14	22	22	22	22	22	22	22	22	22	21	22
p								<.05	<.001	<.001	<.02	
4 Days												
$\bar{X}$	908	136.3	41.8	28.0	15.79	92.2	748.2	9.04	1.09	.229	14.80	26.88
SEM	121	14.2	4.1	1.8	1.85	10.1	67.7	1.02	.09	.019	2.19	2.66
N	13	19	19	19	19	18	19	19	19	19	19	19
p									<.001	<.001		

* Ketosteroids
** Ketogenic steroids

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13. ABSTRACT

Urinary minerals, electrolytes, nitrogen metabolites and steroids were measured in Navy divers following exposure to air at 2 and 7 ATA for 45 minutes.

Total urinary excretion of osmoles, sodium, potassium, calcium, phosphorus, urea nitrogen, uric acid, creatinine, keto- and ketogenic steroids were depressed below control values during the first day following exposure to either 2 or 7 ATA. The mineral and electrolyte changes are related to reduction in urine volume which may result from an anti-diuretic hormone (ADH) mediated response to a hypovolemia induced by dysbaric stress. Although a general rebound effect began to occur by the second post-dive day, a continued depression of uric acid, creatinine, and ketosteroids throughout four days of observation suggests a prolonged period of recovery from diving.

Increasing excretion of hydroxyproline for several days by those men exposed to 7 ATA, implies a latent response in the metabolism of cartilage and bone to this stress.

14. KEY WORDS	LINK A		LINK B		LINK C	
	ROLE	WT	ROLE	WT	ROLE	WT
Mineral metabolism						
Dysbaric stress						
Urinary metabolites						
Steroid excretion						